Effect of Chemical Conditioning of Alfalfa on Drying Rate and Nutrient Digestion in Ruminants[1]

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ABSTRACT

Solutions of K_2CO_3 or KOH were sprayed on freshly cut, third harvest alfalfa. Drying rates of treated alfalfa were faster than control in both laboratory and field trials. Two 3 × 3 Latin square digestion trials were conducted with sheep and dairy cows to evaluate the effects of chemical treatment on digestion of field dried alfalfa hay. Forage composition and DM intake of hays were not influenced by treatment in either sheep or cows. Apparent digestibility of dietary constituents did not differ between control and treated hay in the sheep trial, except for decreased digestibility of NDF with hay treated with KOH. However, digestibility of DM, CP, NDF, and ADF was improved in cows with potassium carbonate treatment of hay. Potassium hydroxide treatment only increased ADF digestion in cows. Extent of NDF digestion in vitro was increased at 12, 24, and 48 h of incubation with both chemical treatments; both treatments decreased the proportion of indigestible NDF. When incubated in situ, particle-associated carboxymethylcellulase activity was greater with potassium carbonate treatment of field dried hay. Treatment with either KOH or K_2CO_3 increased drying rate of alfalfa hay, but only potassium carbonate treatment improved nutrient digestibility.

Received September 28, 1987.
Accepted March 18, 1988.

[1]Mention of commercial products in this paper does not constitute endorsement by the USDA or the ARS.

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INTRODUCTION

Reduction in forage quality is directly influenced by length of field curing during harvesting of alfalfa. Losses result from plant respiration, rain damage, and leaf shatter and are as high as 4% of yield per day (28). Hence, losses during hay harvesting can be minimized by reducing field curing time.

Potassium carbonate increased the drying rate of alfalfa (23, 24), possibly because K_2CO_3 interfered with the hydrophobic properties of cuticular waxes (23). Rotz and Thomas (16) treated alfalfa with Na_2CO_3 or K_2CO_3, alone or in combination with various amounts of methyl esters of long-chain fatty acids, and found that K_2CO_3 was generally more effective in hastening drying of alfalfa. They also reported that chemical conditioning was much more effective on legumes than grasses. Rotz and Davis (14) found that chemical conditioning improved drying rates of all cuttings of alfalfa when drying conditions and application rates were adequate, but mechanical conditioning increased drying rate of only the first cutting. The optimal application rate of drying agents remains controversial (16).

Although drying rate is of major concern, limited research suggests that chemical conditioning may improve forage quality. Chemically treated alfalfa contained more CP and had higher CP and NDF digestibility than untreated hay (5). The purpose of this study was to compare both laboratory and field drying rates of chemically conditioned alfalfa and to evaluate the effectiveness of chemical conditioning on nutrient composition, intake, and digestibility in ruminants.

MATERIALS AND METHODS

Drying Rate Trials

Solutions of either K_2CO_3 or KOH were sprayed on third crop alfalfa (late bud stage) at

cutting using a front mower/conditioner-mounted spray system. Chemicals were applied at 18.0 (K_2CO_3) and 14.6 (KOH) kg/ha; solutions were applied at 280 L/ha. Alfalfa was cut and treated in alternate windrows; the order of cutting and treating was K_2CO_3, then KOH, and finally untreated control. Alfalfa laid in full-width swaths and was field dried for 3 d before raking and baling; hay was lightly rained on once during d 2. Dry matter measurements were made on samples collected from the swaths of each treatment immediately after cutting (zero-h), and collected three times/d (1100, 1400, and 1700 h) on d 2 and 3 after cutting. Three samples from each treatment at each time were collected in bags using a front mounted flail pick-up harvester. Samples were weighed immediately and after drying at 60°C for 72 h.

Field drying rates were estimated using a single exponential model (11): $M/M_0 = e^{-kt}$, where M = moisture content (g H_2O/g DM) at time t (h), M_0 is the initial moisture content (g H_2O/g DM) at t = 0, and k is the fractional drying rate (h^{-1}).

For measuring the drying rate of alfalfa under laboratory conditions, fresh alfalfa grown in the same field was cut prior to field application of chemicals. Alfalfa then was brought to the laboratory where 10 bunches of 20 stems for each treatment were assembled. Each bunch was sprayed evenly by hand with approximately 100 ml of each solution used in field drying studies. These bunches were suspended in the laboratory to dry under ambient temperature and humidity. Bunches were weighed at 3, 6, 20, 30, 44, 68, 92, 116, 140, and 196 h. At 196 h, each bunch was carefully wrapped with cheesecloth to prevent leaf loss and dried at 60°C for 48 h. Laboratory drying data were fit to the biexponential model described by Jones (10):

$$y = a_1 e^{-k_1 t} + a_2 e^{-k_2 t} + a_0$$

where $y_0 = a_1 + a_2 + a_0$ = initial moisture content, y = fraction of initial moisture content (g H_2O/g DM) remaining after time t, a_0 = equilibrium moisture content (g H_2O/g DM), a_1 = fraction of initial moisture (g H_2O/g DM) lost at rapid rate k_1, a_2 = fraction of initial moisture (g H_2O/g DM) lost at slow rate k_2, t = drying time (h), and k_1 and k_2 = fractional drying rates (h^{-1}) for a_1 and a_2.

In Vivo Digestion Trials

Six mature crossbred wethers weighing an average 44.2 kg were placed in individual metabolism crates 3 d prior to beginning the experiment and fed only alfalfa hay. Wethers were randomly assigned to three treatments in a replicated 3 × 3 Latin square. Each period consisted of a 9-d adjustment phase followed by a 5-d collection phase. Only experimental hays (2.0 cm theoretical length of chop) were fed throughout each 14-d period. Total tract digestibility was measured by total collection and using the acid detergent lignin (ADL) ratio technique, assuming ADL is indigestible. Experimental hays were fed ad libitum once daily in amounts to provide 5 to 10% refusals throughout the study. Water was offered twice daily and mineral salt blocks were available ad libitum. Feed intake, corrected for refusals, was recorded once daily. Total daily fecal output was weighed once daily during the last 5 d of each period and a 20% subsample taken. Fecal samples were stored at −20°C, then freeze dried, ground through a 1-mm screen, and composited for analysis. Hay samples and orts were collected daily throughout each 14-d period of the trial. Portions of all hay samples and orts were dried at 60°C for 72 h, ground, composited, and stored for later analysis.

Dry matter was determined at 105°C (1) and N was determined by the Kjeldahl method (1) except that a copper catalyst (Kjeltabs, Tecator Inc., Herndon, VA) was used during digestion. Neutral detergent fiber, ADF, and ADL were measured by the methods of Goering and Van Soest (7) as modified (17). Determination of ADF was not sequential to NDF.

Six Holstein cows, including three with rumen cannulae, which were either nonlactating or in late lactation, were used in a digestibility trial of the same experimental design as the sheep digestion trial. One ruminally cannulated cow was assigned to each treatment. Cows were housed in a stanchion barn; mangers had individual hay racks and enough hay was offered twice daily to assure a 5% refusal. Water and mineral salt blocks were available ad libitum. Each period included a 11-d adjustment followed by a 3-d collection phase. Feed

refusals were recorded once daily prior to the morning feeding. Hay core samples were obtained from each bale of experimental hay throughout the trial. Other procedures for sampling, sample storage, and analyses were those described in sheep digestion trial; total tract digestibility was measured by the ADL ratio technique. Rumen contents were transferred between cannulated cows when diets were switched to reduce time required for adaptation to diets. Rumen fluid was taken from cannulated cows at 0700 and 1000 h on the last day of each period. The pH was measured immediately after sampling. Rumen fluid was preserved (6) and stored at −20°C until later analysis for ammonia (3) and individual and total VFA by gas chromatography (13).

In Vitro Digestion and In Situ Cellulase Activity

In vitro digestion of NDF was determined on laboratory dried alfalfa hays (ground through a 1-mm screen) using the procedure of Goering and Van Soest (7) as modified (17). Whole rumen contents were obtained from a ruminally cannulated, nonlactating Jersey cow fed only alfalfa hay. Rumen fluid inoculum, enriched with particle-associated microorganisms, was prepared by the method of Craig et al. (4). Approximately .5 g of DM was added to each in vitro tube. Tubes were incubated at 39°C for 12, 24, 48, and 72 h, treated with 1 ml saturated $HgCl_2$, then frozen for later analysis. Three incubations were run with triplicate blank and sample tubes for each time point within each incubation. Duplicate samples of field dried control and treated hays were weighed into 100% dacron polyester (R102 Marvelaire White, N. Erlanger, Blumgardt & Co., Inc., 1450 Broadway, New York, NY) bags (60 mm × 100 mm, 52-μm pores). These in situ bags were suspended for 3, 6, 12, 24, 48, and 72 h in the rumen of a ruminally cannulated Jersey cow fed only alfalfa hay (18). Immediately after removal from the rumen, bags were washed in cold water until rinsings were clear, then squeezed thoroughly by hand. After washing, 1 g of wet residue from each bag was dried at 60°C for 48 h for DM determination. Wet residue also was extracted for measurement of particle-associated carboxymethylcellulase (CMCase) activity using a procedure similar to that of Silva et al. (19). With this method, approximately 1 g of wet residue from each bag was transferred to 50 ml centrifuge tubes and mixed with 20 ml of .01 *M* sodium phosphate buffer (pH 6.8) containing lysozyme (20 mg/ml) and treated with 2.5 ml CCl_4 (9). Tubes were vortexed thoroughly for 1 min, then incubated at 39°C for 3 h. Extracted CMCase activity was measured as described by Groleau and Forsberg (8).

Drying rates, in vitro digestibility, and CMCase activity were statistically analyzed as completely randomized designs using one-way ANOVA (21). Intake and digestibility data from sheep and cows were analyzed as a 3 × 3 Latin square replicated two times, and rumen

TABLE 1. Chemical composition of field-treated alfalfa hay.

Item	Treatment			SE
	Control	K_2CO_3	KOH	
DM, %	89.3	89.7	89.8	.24
	(% of DM)			
CP	19.1	18.9	18.6	.58
NDF	42.6	42.5	42.0	.88
ADF	31.7	33.1	32.0	1.39
Acid detergent lignin	7.5	7.3	7.6	.40
Ash	9.6	9.3	9.4	.22
Potassium[1]	2.90	2.94	2.74	...

[1] Potassium concentration from single analyses; therefore, SE were not computed. Potassium contents of laboratory-treated hays were 2.65, 3.74, and 3.80% for control, K_2CO_3, and KOH treatments.

data were analyzed as a single 3 × 3 Latin square (21). Where significant F-values were detected due to diet, mean separation was by least significant differences.

RESULTS AND DISCUSSION

Hay Composition and Drying Rates

Field treatment of alfalfa hay with K_2CO_3 or KOH did not influence the concentration of CP, NDF, ADF, and ADL (Table 1). Concentration of NDF in laboratory dried hay tended to decrease with drying agent treatment (43.2% for control; 40.3% for K_2CO_3; and 41.0% for KOH treated hays, respectively). Treated hay contained more K than control hay in the laboratory trial in contrast to the field trial. This suggests that some K ion may have been lost when field-dried hay was rained on. A series of experiments conducted with alfalfa hay treated with carbonate-salt drying agents (5) indicated no differences in CP and fiber concentration between control and treated hays in one study, but showed that treated hay had significantly higher CP concentration in a second study. Nocek et al. (12) reported CP tended to be higher for untreated control than hay treated with alkaline carbonates and dispersing agents, but NDF and ADF also tended to be lower in treated hays. Losses of DM and CP during field drying have been directly related to length of field exposure (15). Because chemical conditioning reduces the time needed for moisture content to reach a safe concentration for storage, chemical conditioning would be expected also reduce losses of total DM and nutrients. Chemical composition of laboratory dried hay was not affected by drying agent treatment.

Chemical conditioning increased alfalfa drying rate in both laboratory and field trials (Tables 2 and 3, respectively). In the laboratory trial, drying proceeded in two phases (10): 1) A rapid phase with water loss predominantly described by $a_1e^{-k_1t}$, followed by 2) a slow phase with water loss predominantly described by $a_2e^{-k_2t}$ (Figure 1).

Both chemically treated hays dried much faster than control (Table 2), and the rapid drying fraction (a_1) accounted for a greater proportion of total water loss. A greater proportion of total water disappeared from the slow drying fraction (a_2) in control hay. Time from cutting to 60 and 80% DM was significantly reduced for treated hays. Time to dry to 80% DM, a suitable value for storage as hay, was more than twice as long for control (109 h) than treated alfalfa (47 h for K_2CO_3 and 46 h for KOH). It is difficult to account biologically for the rapid and slow drying fractions. These fractions may reflect the relative ease with which water is lost from intercellular spaces and conducting vessels compared with loss from cell sap and cytoplasm (10). Water is lost from the plant by evaporation from sites on the plant

TABLE 2. Effect of drying agents on parameters describing biexponential moisture loss and drying rates of laboratory treated alfalfa hay.

Item[1]	Treatment			SE
	Control	K_2CO_3	KOH	
Y_0, g H_2O/g DM	3.35	3.04	3.10	.23
a_1, g H_2O/g DM	.398[b]	.773[a]	.815[a]	.189
a_2, g H_2O/g DM	.566[a]	.192[b]	.153[b]	.187
k_1, h^{-1}	.098[b]	.125[a]	.136[a]	.018
k_2, h^{-1}	.023	.026	.025	.006
Time to reach 60% DM[2], h	14.8[a]	8.1[b]	8.0[b]	4.2
Time to reach 80% DM[2], h	109.4[a]	46.8[b]	45.5[b]	32.4

[a,b]Means in the same row with different superscripts differ ($P<.05$).

[1] Y_0 = Initial moisture content (t = 0 h), a_1 = the moisture lost at rate k_1, a_2 = the moisture lost at rate k_2, and k_1 and k_2 = the exponential drying rate constants for a_1 and a_2, respectively.

[2] Time computed to be required for alfalfa to dry to 60 and 80% DM (10).

TABLE 3. Effect of drying agents on initial moisture content and drying rates of field-treated alfalfa hay.

Item	Treatment			SE
	Control	K_2CO_3	KOH	
Initial moisture content, %	79.3	77.1	76.1	1.80
Drying rate, h^{-1}	.029[b]	.039[a]	.037[a]	.005
Time to reach 60% DM[1], h	31.6[a]	23.5[b]	24.8[b]	3.69
Time to reach 80% DM[1], h	55.5[a]	41.3[b]	43.5[b]	6.72

[a,b]Means in the same row with different superscripts differ ($P<.05$).

[1]Time computed to be required for alfalfa to dry to 60 and 80% DM (11).

surface. A gradient of water vapor is maintained between the surface and the surrounding atmosphere; the ease with which vapor moves away from the plant influences the drying rate (10). Water vapor is greatest near the plant surface. Drying agent treatment may lower resistance to diffusion of water from the surface due to loss of hydrophobicity of the cuticular layer (23).

Unlike the laboratory trial, the drying data from field cured hay fit a single exponential (11), possibly because only six time points were used. Alfalfa drying rates also were significantly enhanced with either drying agent (Table 3). Average field drying time to reach 80% DM was shortened by 12 h with KOH and 14 h with K_2CO_3. Several studies have investigated the use of Na_2CO_3 or K_2CO_3 based solutions to improve drying rate. Treating alfalfa with aqueous K_2CO_3 reduced drying time in laboratory (23) and field trials (24).

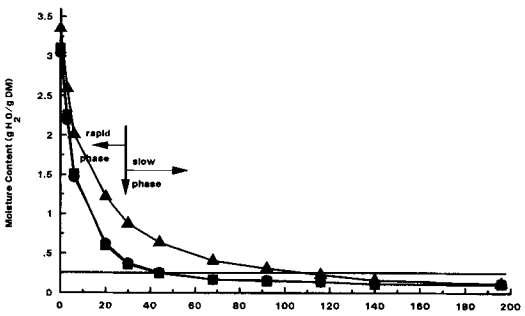

Figure 1. Changes in moisture content of control (▲), potassium carbonate-treated (●), and potassium hydroxide-treated (■) alfalfa hay dried under laboratory conditions.

In Vivo Digestion Trials

Intake and total tract digestibility in sheep fed field dried hay are in Table 4. Drying agent treatments did not affect DM intake (DMI), or digestibility of DM, CP and ADF. Only NDF digestibility in KOH treated hay was significantly decreased compared with control and K_2CO_3 treatment. This effect appears spurious because ADF digestibility was not changed, and in vitro digestibility of NDF actually increased with KOH in laboratory treated hay (Table 5). Total collection and ADL ratio techniques yielded essentially identical estimates of digestibility (Table 4). This indicated complete ADL recovery, and an ADL ratio technique was used to determine in vivo digestibility in dairy cows. Ehle et al. (5) reported digestibilities, based on lignin ratio, which were higher than those from total collections.

Intakes of DM and digestibility determinations (by ADL ratio) in dairy cows fed field dried hay are in Table 6. Drying agent treatment of alfalfa caused no differences in DMI. However, digestibilities of DM, CP, NDF, and ADF were significantly enhanced by K_2CO_3 treatment. Only ADF digestibility was increased with KOH treatment compared with digestibility of control hay.

There has been limited work on the effects of chemical conditioning on forage nutritive value. The improvements in digestibility of alfalfa treated with K_2CO_3 or KOH in this study may be due to modification of the alfalfa cuticle, which is thought to be indigestible by rumen microorganisms (2, 26). These chemical treatments also may alter the physical and chemical distribution of plant nutrients and finally may affect ruminal digestion of fibrous components by allowing greater accessibility of

TABLE 4. Effect of drying agent treatment of alfalfa hay on DM intake and total tract digestibilities in sheep.

Item	Treatment			SE
	Control	K_2CO_3	KOH	
DM Intake, kg/d	1.36	1.43	1.39	.22
Digestibility (total collection), %				
DM	55.4	55.6	55.3	.39
CP	67.8	67.7	68.4	2.30
NDF	40.5[a]	41.3[a]	33.1[b]	1.20
ADF	36.2	37.3	35.0	2.53
Digestibility (ADL[1] ratio), %				
DM	56.2	54.9	56.2	1.69
CP	68.5	67.3	69.3	2.76
NDF	41.6[a]	40.0[a]	34.7[b]	4.19
ADF	38.4	36.2	37.2	3.31

[a,b]Means in the same row with different superscripts differ ($P<.05$).

[1] ADL = Acid detergent lignin.

TABLE 5. Extent of in vitro digestion of NDF for laboratory-treated alfalfa hay.

Incubation time	Treatment			SE
	Control	K_2CO_3	KOH	
(h)	(% of total NDF)			
12	24.9[b]	28.2[a]	29.9[a]	.43
24	35.4[b]	39.6[a]	40.7[a]	1.20
48	40.6[b]	45.3[a]	47.4[a]	1.00
72	42.5[b]	49.0[a]	49.0[a]	.77

[a,b]Means in the same row with different superscripts differ ($P<.05$).

TABLE 6. Effect of drying agent treatment of alfalfa hay on DM intake and total tract digestibilities in dairy cows.

Item	Treatment			SE
	Control	K_2CO_3	KOH	
DM Intake, kg/d	15.7	15.1	14.9	.28
Digestibility,[1] %				
DM	58.9[b]	61.9[a]	60.0[ab]	.79
CP	66.9[b]	70.6[a]	68.4[ab]	.90
NDF	43.3[b]	47.1[a]	44.3[b]	.93
ADF	40.9[b]	44.1[a]	44.9[a]	1.10

[a,b]Means in the same row with different superscripts differ ($P<.05$).

[1] Digestibilities were calculated from acid detergent lignin ratio.

microorganisms to plant tissue (12). Weighart et al. (27) treated alfalfa hay in the laboratory with K_2CO_3, alone or in combination with surfactants, and observed no differences in DM digestibility in vitro. Valentine et al. (25) showed similar results with K_2CO_3 treatment of alfalfa hay. Nocek et al. (12) applied commercial drying agents, with or without surfactants, to first-cutting alfalfa at two different concentrations. Although there were some changes in rates of NDF and ADF digestion in situ, those workers (12) concluded that, regardless of surfactant and drying agent concentration, treating alfalfa hay with drying agents did not increase overall NDF and ADF digestion. These findings differ from those of Ehle et al. (5) who reported that intake and digestibility of CP and NDF in sheep were significantly higher for alfalfa hay treated with carbonate salt-based drying agents. Differences in digestibility between sheep and dairy cows are difficult to explain. Both chemical treatments had similar affects on alfalfa drying rate, but only K_2CO_3 was also effective on in vivo digestion in cows.

Rumen pH and most VFA were not affected in cows fed hay treated with drying agents (Table 7). However, propionic acid decreased, and butyric acid increased for the cows fed K_2CO_3-treated hay. Lower rumen ammonia concentration in cows fed K_2CO_3-treated hay may be related to increased DM and fiber digestibility (Table 6). Increased digestion would be expected to stimulate ammonia uptake for microbial growth, thereby lowering ruminal ammonia concentration.

In Vitro Digestion and In Situ Cellulase Activity

Extents of in vitro NDF digestion of laboratory dried hays are in Table 5. Digestion of NDF was significantly greater at all times for hay treated with K_2CO_3 or KOH. Indigestible NDF (72 h) in control, K_2CO_3-treated, and KOH-treated hay was 57.5, 51.0, and 51.0%, respectively, indicating both treated hays contained more digestible NDF than control, despite their origin from the same material. Craig et al. (4) reported a linear increase in rate of in vitro digestion of potentially digestible NDF in alfalfa treated with graded amounts of a drying agent containing primarily K_2CO_3, when the in vitro inoculum was enriched with particle-associated rumen microorganisms.

Field dried hay treated with K_2CO_3, when incubated in the rumen in situ, had higher particle-associated CMCase activity at 6 and 12 h (Figure 2). Activity of CMCase was maximal at 6 h of incubation regardless of treatment, and declined rapidly thereafter; enzyme activity was similar between KOH treated hay and control at all incubation times. Higher CMCase activity reflected increased NDF digestion of K_2CO_3-treated hay in vitro and in the cow trial. Increased cellulolytic enzyme activity is probably due to greater adhesion of cellulolytic

TABLE 7. Effect of feeding alfalfa hay treated with drying agents on ruminal pH, ammonia, and VFA in dairy cows.

Item	Treatment			SE
	Control	K_2CO_3	KOH	
pH	7.05	7.10	7.10	.10
Ammonia N, mg/dl	26.3[a]	22.4[b]	24.5[ab]	.92
Total VFA, m*M*	97.1	98.0	95.0	4.57
Acetate, molar %	70.1	70.2	70.0	.29
Propionate, molar %	15.6[ab]	14.9[b]	15.8[a]	.27
Butyrate, molar %	8.6[b]	9.4[a]	8.6[b]	.05
Isobutyrate, molar %	1.85	1.85	1.78	.04
Isovalerate, molar %	2.60	2.58	2.55	.06
Valerate, molar %	1.30	1.18	1.33	.06
Acetate:propionate	4.51	4.73	4.45	.10

[a,b]Means in the same row with different superscripts differ ($P<.05$).

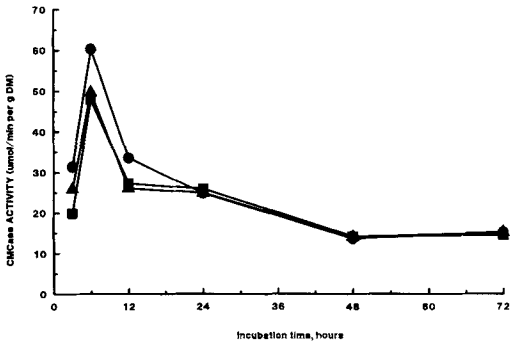

Figure 2. Particle-associated carboxymethylcellulase (CMCase) activity obtained when control (▲), potassium carbonate (●), and potassium hydroxide (■) field-treated alfalfa hay were incubated in situ in the rumen.

bacteria on the surface of hay cell walls; this may be a major factor accounting for increased NDF digestibility in K_2CO_3-treated hay. Cellulolytic bacteria must be present in large numbers and must be in close contact with the plant cell wall to saturate the exposed substrate surface with cellulolytic enzymes (9, 20) and to obtain maximal rates of cell wall digestion (22). Examination of ruminally incubated straw by electron microscopy indicated that the plant surface layers formed barriers to microbial digestion (22). Removal of alfalfa surface cutin layer by drying agent treatment may possibly explain the increased CMCase activity and NDF digestibility observed with K_2CO_3-treated alfalfa hay.

CONCLUSIONS

Both K_2CO_3 and KOH treatment increased drying rates of third cutting alfalfa hay. Therefore, chemical conditioning may reduce the interval from cutting to baling and decrease the risk of quality reduction from adverse weather. Potassium carbonate was more effective than KOH in enhancing alfalfa fiber digestion in vivo. Although no positive effects on digestion were observed with either chemical in sheep, K_2CO_3 increased digestibility of CP, DM, and fibrous components of alfalfa in dairy cows. Only K_2CO_3 increased in vitro fiber digestibility and in situ particle-associated cellulase activity.

ACKNOWLEDGMENTS

The authors gratefully acknowledge the excellent technical assistance of Brad Ricker, Heidi Mier, Greg Stumpf, and Jeff Jackson during the course of this work.

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